Claims

- Method for increasing the production of 1. cysteine, glutathione and methionine, and of sulphur derivatives thereof, by plant cells and plants, the 5 said method consisting in overexpressing an SAT in plant cells and plants containing the said plant cells.
 - Method according to claim /, characterized in that the SAT which is overexpressed in plant cells is a cysteine-sensitive SAT.

- Method according to claim 2, characterized in that the SAT is a plant SAT or a native SAT of bacterial origin.
- Method according to claim 1, characterized in that the SAT which is overexpressed in plant cells is a cysteine-insensitive SAT.
- Method according to claim 4, characterized in that the SAT is a plant SAT or an SAT of bacterial origin, or a mutated plant SAT, rendered cysteine-insensitive by mutagenesis.

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- Method according to one 5, characterized in that the SAT is overexpressed in the cytoplasm of plant cells.
 - Method according to claim 6, characterized in that the SAT is an SAT of bacterial origin.
 - Method according to claim 6, characterized in that the AT is a plant cytoplasmic SAT, in particular from Arabidopsis thaliana.

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Method according to claim 8, 9. characterized in that the SAT is SAT3 which is epresented by SEQ ID NO 1.

- 10. Method according to claim 6,
- characterized in that the SAT is a non-cytoplasmic plant SAT from which has been removed its signal(s) for addressing to cellular compartments other than the cytoplasm.
 - Method according to claim 10,
- 10 characterized in that the SAT is SAT1' which is represented by SEQ ID NO 2.
 - 12. Method according to one of -5, characterized in that the SAT is overexpressed in mitochondria.
 - Method according to claim 12, 13. characterized in that the SAT is overexpressed in the cytoplasm in the form of a signal peptide/SAT fusion protein, the mature functional SAT being released inside mitochondria.
- 14. Method according to claim 13, 20 characterized in that the mitochendrial addressing signal peptide consists of at least one signal peptide from a natural plant protein which is located in mitochondria, such as for example, the SAT1 signal peptide which is represented by amino acids 1 to 63 in SEQ ID NO 3

- Method according to claim 13, 15. characterized in that the SAT is a mitochondrial SAT of plant origin, in particular from Arabidopsis thaliana.
 - 16. Method according to claim 15,
- 5 characterized in that the SAT is SAT1 which is represented by SEQ ID NO 3.
 - Method according to claim 6 characterized in that the SAT is overexpressed in chloroplasts of plant cells.
 - 18. Method according to Llaim 17, characterized in that the SAT is overexpressed in chloroplasts by integration, into chloroplast DNA of plant cells, of a chimeric gene comprising a DNA sequence encoding the said SAT, under the control of 5' and of 3' regulatory exements which are functional in chldroplasts.
 - Method according to claim 17, 19. characterized in that the SAT is overexpressed in the cytoplasm in the form of a transit peptide/SAT fusion protein, the mature functional SAT being released inside chloroplasts.
 - 20. Method according to claim 19, characterized in that the SAT is homologous with the transit peptide.
- Method according to claim 20, 25 characterized in that the SAT is a chloroplast SAT of plant origin, in particular from Arabidopsis thaliana.

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22. Method according to claim 21, characterized in that the SAT is SAT2 or SAT4 which are represented by SEQ ID NO 5 or NO 6, respectively.

Method according to claim 19/

characterized in that the SAT is heterologous with the transit peptide.

Method according to claim 13,

characterized in that the SAT is a cytoplasmic SAT of plant origin or an SAT of bacterial origin, as defined in one of claims 3 to 5 or 9 to 11.

Method according to either of claims and 24, characterized in that the transit peptide is a transit peptide from another protein which is located in plastids.

15 26. Method according to claim 25, characterized in that the transit peptide consists of a plant EPSPS transit peptide or a plant RuBisCO ssu transit peptide.

Method according to cither of 27. 20 -and 26, characterized in that the transix peptide comprises a transit peptide from a plant protein which is located in plastids, and, between the C-terminal portion of the transit peptide and the N-terminal portion of the SAT, a portion of sequence from the 25 mature N-terminal region of a protein which is located in plastids.

Method according to claim 27, characterized in that the portion of sequence comprises

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generally less than 40 amino acids from the N-terminal portion of the mature protein, preferably less than 30 amino acids, more preferably between 15 and 25 amino acids.

29. Method according to either of claims 27

and 28, characterized in that the transit peptide
comprises, between the C-terminal portion of the
N-terminal portion of the mature protein and the
N-terminal portion of the SAT, a second transit peptide
from a plant protein which is located in plastids.

30. Method according to claim 29, characterized in that the transit peptide is an optimized transit peptide (OTP) made by fusing a first transit peptide with a portion of sequence from the mature N-terminal region of a protein located in plastids, which is fused with a second transit peptide.

31. Transit peptide/SAT fusion protein, characterized in that the SAT is heterologous with the transit peptide.

32. Fusion protein according to claim 31, as defined in claims 24 to 30.

33. Nucleic acid sequence encoding a transit chi.m 3/peptide/SAT fusion protein according to either of elaims 31 and 32.

34. Chimeric gene comprising a coding sequence as well as heterologous 5' and 3' regulatory sequences, which are able to function in a host organism, characterized in that the coding sequence

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comprises at least one nucleic acid sequence which encodes an SAT.

- 35. Chimeric gene according to claim 34, characterized in that the host organism is chosen from bacteria, for example *E. coli*, yeasts, in particular of the genera *Saccharomyces*, *Kluyveromyces* or *Pichia*, fungi, in particular *Aspergillus*, baculoviruses, or plant cells and plants.
- 36. Chimeric gene according to claim 35,
 10 characterized in that the host organism is a plant cell or a plant which contains it .
- 37. Chimeric gene according to claim 36, characterized in that the 5' regulatory element comprises regulatory sequences which are promoters in plant cells and plants, and are chosen from promoters which are expressed in plant leaves, constitutive promoters, or light-dependent promoters of bacterial, viral or plant origin.
- 38. Chimeric gene according to claim 36,
 20 characterized in that the 5' regulatory element
 comprises regulatory sequences which are promoters in
 plant cells and plants, and are chosen from seedspecific promoters.
- 39. Chimeric gene according to claim 38,
 25 characterized in that the promoter is chosen from the promoters for napin, phaseolin, glutenin, zein,
 helianthinin, albumin and oleosin.

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40. Chimeric gene according to one of claims 34 to 39, characterized in that the nucleic acid sequence which encodes an SAT encodes an SAT as defined in claims 2 to 30.

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41. Chimeric gene according to one of claims 84 to 39, characterized in that the nucleic acid sequence which encodes an SAT is the nucleic acid sequence according to claim 33.

42. Cloning and/or expression vector for transforming a host organism, characterized in that it contains at least one chimeric gene as defined closw 34 according to one of claims 34 to 41.

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43. Method of transforming host organisms, characterized in that at least one nucleic acid sequence according to claim 33, or a chimeric gene according to one of claims 34 to 41, is integrated into the genome of the said host organism.

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of the vector according to claim 42.

45. Method according to either of claims 43

and 44, characterized in that the host organism is chosen from bacteria, for example *E. coli*, yeasts, in particular of the genera *Saccharomyces*, *Kluyveromyces* or *Pichia*, fungi, in particular *Aspergillus*,

- 25 baculoviruses, or plant cells and plants.
 - 46. Method according to claim 45, characterized in that the host organism is a plant cell or a plant which contains it.

- 47. Method according to claim 46, characterized in that the plant is regenerated from a transformed plant cell.
 - Method according to claim 47,
- characterized in that the host organism is a monocotyledonous plant, in particular chosen from cereals, sugar cane, rice and maize, or a dicotyledonous plant, in particular chosen from tobacco, soybean, rape, cotton, beet and clover.
- Transformed host organism, characterized 10_ in that it comprises at least one nucleic acid sequence according to claim 33, or a chimeric gene according to one of claims 34 to 41.
 - Host organiam according to claim 49, $^\prime$ characterized in that it is δ ptained by the method according to one of claims 43 $\sqrt{6}$ 48.
 - Plant cell, characterized in that it 51. comprises at least one nucleic acid sequence according to claim 33, or a chimeric gene according to one of claims 34 to 41.
 - Genetically modified plant, characterized in that it comprises at least one plant cell according to claim 51.
- Plant according to claim 52, /characterized in that the phant is regenerated from a 25 plant cell according to claim \51.
 - 54. Genetically modified plant, characterized in that it is derived from the culture

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and/or crossing of regenerated plants, according to claim 53.

- 55. Genetically modified plant according to Caim 52
 one of claims 52 to 54, characterized in that it is a
- 5 monocotyledonous plant, in particular chosen from cereals, sugar cane, rice and maize, or a dicotyledonous plant, in particular chosen from tobacco, soybean, rape, cotton, beet and clover.
- 56. Genetically modified plant according to Uum 52

 10 one of claims 52 to 55, characterized in that it comprises other genes of interest.
- 57. Genetically modified plant according to claim 56, characterized in that it comprises at least one other gene which modifies the content and quality of the proteins of the said plant, in particular in the leaves and/or seeds.
 - 58. Genetically modified plant according to either of claims 56 and 57, characterized in that the gene encodes a protein enriched in sulphur-containing amino acids.
 - 59. Seeds of genetically modified plants according to $\frac{52}{\text{one of claims }52}$ to $\frac{58}{\text{one of claims }52}$

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